IMPROVED AEROSOL METHODOLOGY FOR APPLYING CPT TO CONTROL ROOSTING SPECIES OF PEST BIRDS

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ABSTRACT: Colony roosting species of blackbirds (Icterinae), including European starlings (*Sturnus vulgaris*), can be significant pests. The chemical avicide CPT has been applied experimentally in the U.S. and France to roosting blackbirds. However, apparent effective aerial application rates are high, 44-101 kg/ha (40-90 lbs/a), and the actual efficacy, expressed as percent mortality, is difficult to determine. We assessed CPT as a respirable aerosol for potential use as a roost avicide. Starlings exposed to 17 ppm CPT for 5.5 min received lethal doses. The birds appear very sensitive to CPT administered in this manner. The methods of CPT entry into the birds include respiratory, ocular and dermal. It is proposed that a field application rate of 3.4-5.6 kg of CPT per hectare (3-5 lbs/a) would be effective.

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INTRODUCTION

Colony roosts of blackbirds (Icterinae), including European starlings (*Sturnus vulgaris*), can pose serious problems as pests (Caccamise 1990, Heisterberg et al. 1990). The same roosting birds often cause agricultural damage or nuisance situations at locations away from the roost. Congregation of these birds into roosts, however, presents an opportunity to apply a lethal agent for population reduction. In the last decade, the chemical avicide CPT (3-chloro-4-methylbenzenamine, DRC-1347) has received serious attention as a possible roost toxicant. However, when CPT was applied aerially at 44-101 kg/ha (40-90 lbs/a), mortality estimates were variable, because birds left the roost and dispersed before death. The high application rate also raises concerns about environmental contamination (Douville de Franssu et al. 1988, Heisterberg et al. 1990).

The authors hypothesized that a respirable aerosol of CPT (particles < 10µm diameter) would both kill a high percent of blackbirds at a roost and leave a low level of residue at the application site and surrounding vicinity. Previously tested field applications contained little or no CPT in a respirable form. We decided to test the concept using starlings in the controlled environment of a laboratory equipped with aerosol generation and measurement instrumentation.

METHODS

The exposure system was established in a testing room with an exhaust system that kept the room under negative air pressure. Room temperature was 19°-24° C. Starlings were held in another room in individual 24.1 x 17.8 x 17.8 cm wire cages before and after exposure. Commercial bird food and water were freely available and a 12 hr/12 hr room light/dark cycle was used plus ambient outside window light. Holding room temperature was 20°-24° C.

The exposure system consisted of six units: Atmosphere conditioning; avicide aerosol generation; aerosol blending; bird exposure; avicide collection; and atmosphere exhaust. Air flowed through the system due to negative pressure caused by a stainless steel vacuum cleaner unit (Dayton Electrical Mfg. Co., Chicago, IL[1]) attached to the exhaust end. Rate of exhaust flow was controlled with a variable auto transformer (Staco Energy Products, Dayton, OH).

Atmosphere Conditioning

Intake air was humidified with a commercial console humidifier (Emerson Electric Co., St. Louis, MO) with a custom plexiglass humidity-collection chamber (61.0 x 30.5 x 30.5 cm) located over the exhaust of the humidifier at the top of the console. Humidity was regulated manually with a variable auto transformer that controlled the speed of the humidifier. Relative humidity (RH) and temperature of intake air entering the area where the aerosol was generated was monitored with a hygro-thermograph (The Bendix Corp., Baltimore, MD) located in a custom-made plexiglass chamber (62.0 x 31.8 x 32.4 cm). Humidity was maintained between 30% and 50% RH and temperature between 21° and 24° in the intake air.

Filtration occurred after the air left the humidity-collection chamber, by routing the air through a PVC tube to a custom-made plexiglass filter bay (33.0 x 33.0 x 15.2 cm). The collected air then was passed through an absolute filter unit (30.5 x 30.5 x 30.5 cm) (Young and Bertke Co., Cincinnati, OH) with a pleated coarse filter (American Air Filter, Louisville, KY), a charcoal bed and a HEPA filter (Mine Safety Appliances Co., Pittsburgh, PA). The air then flowed through a PVC pipe to the hygro-thermograph chamber previously described.

Avicide Aerosol Generation

The conditioned air then flowed through a PVC tube into a 91.4 x 63.5 x 91.4 cm glove box (Lab-Con Co., Kansas City, MO) containing aerosol generators (nebulizers). The glove box contained a plexiglass tube 96.5 cm long with an inside diameter of 6.4 cm and a series of side ports where up to 10 nebulizers could be attached. Model X70/N nebulizers (Burton Co., Van Nuys, CA) were mounted on jars that contained up to 100 ml of the CPT formulation. The nebulizers were operated at 138 kPa (20 psi) input, breathable air, and the respirable aerosol produced was drawn down the plexiglass tube into a flexible plastic, acid resistant tube exiting the glove box to join a stainless steel inlet tube on the aerosol blending chamber. The aerosol formulation contained by volume 10% CPT, 30% isopropyl alcohol, 23% propylene glycol and 37% water. The formulation was the same as that used in a 1989 roost spray conducted by the Denver Wildlife Research Center (DWRC) in Mississippi (Heisterberg et al. 1990).

[1] Use does not constitute endorsement by the U.S. Government.

Aerosol Blending

The aerosol atmosphere traveled through a stainless steel tube into the top of the stainless steel inhalation chamber (91.4 x 91.4 x 132.1 cm) used to mix the aerosol. As the aerosol entered the chamber at the top, the turbulence created resulted in a mixing action which created a homogeneous atmosphere consisting of air, CPT formulation droplet particles and vaporized formulation components. This aerosol "cloud" was monitored by a custom made aerosol sensor that was an infrared emitting and sensing probe located on the upper side of the chamber (Higgins et al. 1978, Holmberg et al. 1985). This probe, connected to a strip chart recorder, allowed the researcher to monitor the density of the cloud and determine when the atmosphere in the inhalation chamber had reached a steady state of reflectance and thus concentration. This usually occurred about 20 min after aerosol generation began. The temperature of the chamber atmosphere was maintained between 21° and 23° C. During any time birds were not being exposed, bypass valves were open to allow the treated atmosphere to flow through the system but bypass the bird exposure chamber.

Bird Exposure

A small chamber with a flowing atmosphere for the exposure of the birds was selected for several reasons. The small size allows rapid filling with a homogeneously treated atmosphere drawn from the aerosol blending chamber and rapid evacuation. Exposures of realistic duration and known CPT concentration could be made. The chamber achieved a steady concentration in less than 15 sec and emptied in the same time. When birds were in the chamber but not being exposed, valves were opened to allow fresh room air to move through the chamber.

The exposure chamber was constructed with 1.3 cm plexiglass to form a box with inside dimensions of 27.9 x 19.0 x 17.8 cm. The lid was sealed with a rubber gasket and snap lock fasteners. The exposure atmosphere entered and exited the ends of the box through a 1.9 cm circular hole in the center of each end. A 0.5 cm thick 8.9 x 8.9 cm plexiglass baffle plate was attached inside the box 1.6 cm in front of each hole to prevent the flow of atmosphere from directly impacting the birds.

Five minutes before exposure two starlings were placed into a 22.9 x 14.0 x 15.2 cm wire mesh cage, which was placed into the plexiglass exposure chamber. The cage was constructed of 1.3 cm mesh stainless steel wire and had a divider of the same wire mesh in the middle to separate the cage into two equal sized compartments. The birds were exposed to the test atmosphere for 5 min. and after the exposure valve was closed, remained in the chamber for an additional 30 sec during which time the chamber was purged with room air. Even though all CPT atmosphere in the chamber appeared to evacuate in 15 sec, the purge was conducted for 30 sec to be certain the air was free from CPT. The birds were then removed from the exposure chamber and placed back into the holding cages, one bird per cage.

The atmosphere was sampled continuously during the 5.5 min of exposure. The atmosphere was collected by drawing a sample, from a tube inserted into the center of the exposure chamber, through a fiberglass filter pad at the rate of one liter of atmosphere per minute. The 45 mm diameter borosilicate glass filter discs and acrylic filter holders were obtained from Phipps and Bird Co., Richmond, VA. The CPT on each filter pad was extracted by a 10 min. wash with 25 ml of the aerosol formulation (without the CPT) and the quantity of CPT present was determined by spectrophotometry using a CPT standard curve for light transmission at 236 nm. The CPT concentration in the exposure system atmosphere could be controlled within a specific range, mainly by the number of nebulizers used, but the exact concentration in the range could only be determined by the sample collected on the pad during exposure.

The exposure atmosphere was examined for particle size by using a model 02-130 seven stage cascade impactor (Intox Products, Albuquerque, NM). A 5 min atmosphere sample was drawn from the center of the exposure chamber at the rate of one liter per min and through the cascade impactor where the particles were partitioned into seven size categories, with the smallest and last category being vapor. The CPT present in each category was determined by carrier formulation extraction of the collecting discs and spectrophotometry.

Avicide Collection and Atmosphere Exhaust

In both the bypass and exposure modes, the CPT atmosphere was passed through a seven bank, DX-grade coalescing filter (Balston Filter Products, Lexington, MA) to remove the CPT and then exhausted to the outside atmosphere via a flexible tube to the ceiling exhaust in the room.

Test Parameters

Thirty-three starlings were tested. The three exposure treatment groups were a control (formulation minus CPT), and approximate LC_{50} and LC_{100} levels. Treatment levels and related information are presented in Table 1. Two nebulizers were used to generate the control and high dose levels and one to generate the low dose level. Typically, almost all particles were less than 5 µm in diameter and, depending on the number of nebulizers used and relative humidity, often the CPT would have partially vaporized to a gas. The atmosphere flow through the plexiglass exposure chamber was kept at approximately three liters per sec with a temperature of 22° C ± 1°. Since the volume of the chamber was 9.5 liters, without the birds or cage present, the majority of the atmosphere in the chamber would change every three seconds. This is equivalent to a 0.3 kph (0.2 mph) wind travelling 27.9 cm, the inside length of the chamber, in 3.1 sec. The turbulence in the chamber created by the baffle plates resulted in atmosphere

Table 1. Starling CPT respirable aerosol exposure levels.

N	Dose level	Concentration[a,b] (µg/l air)	PPM (v/v)
9	control (carrier only)	0.0	0
14	Low	46.3 ± 4.2	10
10	High	81.4 ± 6.5	17

[a]Mean ± one standard deviation.
[b]The concentrations and ppm values during the first 5 min of exposure actually are slightly higher because the concentration in the exposure chamber was being reduced to zero during the last 30 sec of the 5.5 min exposure.

movement somewhat more rapid than 0.3 kph.

Our impression at the start of the study was that the mode of CPT entry into the birds was mainly by respiration even though the starlings were treated with a whole body exposure. After calculating the volume of air a starling would inhale during an exposure and examining how a low air concentration of CPT killed the initial starlings dosed, it became apparent that it was very unlikely a starling could receive a lethal dose of CPT via respiration alone while in a 17 ppm atmosphere for 5.5 minutes. To further explore mode of entry, 15 starlings were exposed in the same manner as in the main study but had selected portions of their anatomy protected from CPT exposure (Table 2). We questioned if body only exposed birds were being dosed with CPT through preening. To answer this question, two of the body only exposed birds were dosed and then plastic funnel barriers were attached around their necks so preening was prevented but feeding and drinking were possible.

Table 2. CPT mode of absorption in starlings exposed to a respirable aerosol (5 min exposure plus 30 sec venting).

Exposure Mode	N[a]	Dose[b] (µg/l air)	Mortality
Respiration only (bill was exposed)	6	120 ± 34 (82-160)	0/6
Head only	3	103 ± 9 (98-114)	3/3
Head only, eyes covered	2	107.5 ± 4 (105-110)	0/2
Body only	4	96 ± 12 (90-108)	4/4

[a]Males
[b]Mean ± one standard deviation and range.

RESULTS AND DISCUSSION

The intent of the study, to expose starlings to respirable aerosols of CPT that would be lethal to about 50% of the birds at a low dose and to about 100% of the birds at a high dose, was met (Table 3).

As expected, some birds in the lower dose group died later than the birds from the higher dose group. All 10 of the birds in the low dose group died by the end of the third 24 hr period after they were exposed (2 died during the 3rd day). All 10 of the high dose birds died before the start of the 3rd day. Administration of CPT as a respirable aerosol was very toxic to starlings. A comparison with two better known toxic chemicals illustrates this. For carbon monoxide, a 35 ppm exposure for 1 hr is the safe limit for humans (National Research Council, 1977). For hydrogen cyanide, a 150 ppm exposure for 0.5-1.0 hr may be fatal in humans (Budavari 1989). For CPT, a 17 ppm exposure for 5 min is fatal in starlings. Even with the limited number of birds used in the mode of entry probe, it is clear that respiration was not the only mode of entry for respirable CPT in the starling (Table 2). Respiration alone killed no birds even though the mean CPT air concentration of 120 µg/l was almost 50% greater than the mean of 81.4 µg/l for the high dose group in the main part of the study, where 10 of 10 birds died. When head exposure was added to respiration, all three birds died, indicating that absorption by the skin on the head, eyes or both added to the respiratory dose. Covering the eyes resulted in no deaths, indicating that absorption by the eyes is an important part of the total dose received in a head only exposure. Body only exposure was also lethal to all birds even when preening was prevented. To demonstrate that respiration alone could be potentially lethal at high enough levels, a single starling was dosed with the bill only exposed at 199 µg/l; this bird died.

Table 3. Mortality of starlings exposed to a respirable aerosol of CPT.

Dose (µg/l air)	Mortality
0.0	0/9
46.3	10/14
81.4	10/10

Based on results of this study, we believe that the field application rate for CPT might be successfully reduced from 44-101 kg/ha (40-90 lbs/a) to as low as 3.4-5.6 kg/ha (3-5 lbs/a). Four kg of CPT in a respirable aerosol would theoretically form a cloud over one hectare 4.9 m deep at a concentration of 17 ppm. With slight or no wind, birds in the roost would probably be exposed to a 17 ppm concentration of CPT for at least 5 minutes.

If respirable aerosols of CPT are highly toxic to target species (blackbirds) and much less toxic to most non-target species, as is true in oral and dermal CPT dosing, CPT might offer an environmentally acceptable avicide. If avicides prove to be generally unacceptable, aerosols might still have uses for delivery of future nonlethal alternative methods such as immunocontraceptive vaccines.

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